A PATHWAY FOR PLEASANT TOUCH
LINKING PERIPHERAL RECEPTORS TO CENTRAL PROCESSING AND HEDONIC EXPERIENCE

UNIVERSITY OF GOTHENBURG

LINE SOFIE LÖKEN

Department of Neuroscience and Rehabilitation
and Department of Physiology
Institute of Neuroscience and Physiology
at Sahlgrenska Academy
University of Gothenburg
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Line Sofie Löken, Department of Neuroscience and Rehabilitation, Institute of Neuroscience and Physiology, University of Gothenburg, Gothenburg, Sweden, 2009.

Abstract

This thesis investigates the mechanisms underpinning pleasant touch, describes a pathway from peripheral nerve endings in the skin to the insular cortex, and relates these findings to the subjective hedonic experience of touch.

In Paper I, the relationship between primary afferent encoding and perception of pleasantness was investigated by combining microneurography recordings from human mechanoreceptors with psychophysical measurements during soft brush stroking at 6 different velocities between 0.1–30 cm/s. Results showed that low-threshold unmyelinated fibers (C tactile, CT), but not myelinated afferents, responded most vigorously to intermediate brushing velocities (1–10 cm/s), which were perceived by subjects as being the most pleasant.

In Paper II, a group of patients with reduced C fiber density due to a rare inheritable disorder (hereditary sensory and autonomic neuropathy type V, HSAN-V), provided the opportunity to address how pleasantness is perceived when the number of CT afferents is reduced. In comparison with healthy control subjects the C fiber denervated patients displayed atypical pleasantness ratings for soft brush stroking across different brushing velocities. These results suggest that conventional pleasant touch is dependent on CT fiber density.

CT afferents are lacking in glabrous skin which suggests that pleasant touch is perceived differently in the palm compared to the forearm. In Paper III, three different experiments were performed on three different groups of experimentally naive, healthy subjects. In experiment 1, a series of brush strokes was first applied to the palm followed by a series of brush strokes on the arm; in experiment 2, this order was reversed. In experiment 3, brush strokes were applied to the palm and arm in an alternating fashion. In experiment 1 subjects rated gentle stroking as less pleasant on the palm compared to the arm. In experiments 2 and 3, similar ratings were seen for the palm and arm. These results suggest that the perception of pleasantness on the palm is affected by previous stimulation of the arm, but not vice versa. It was speculated that assessment of pleasant touch may be influenced by affective reactions elicited through activation of the CT afferent pathway.

Paper IV investigated whether CT afferents project to the cortex in a somatotopic fashion. In order to distinguish between cortical activations evoked by myelinated (Aβ) fibers and those specifically related to CT afferents six healthy subjects were compared to a unique patient (GL), who lacks Aβ afferents. Soft brush stimulation was applied to the participants' arm and thigh during functional magnetic resonance imaging (fMRI). CT afferents were shown to project somatotopically to the posterior insular cortex in a similar fashion to those previously identified for signalling temperature and pain.

In conclusion, this thesis provides an improvement to the understanding of the neural substrates governing pleasant touch. Further knowledge on the mechanisms behind affective touch may be useful for understanding certain psychiatric disorders, such as autism, where gentle touch is perceived as unpleasant.

Keywords: CT afferent, unmyelinated, pleasant, touch, human, microneurography
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POPULÄRVETENSKAPLIG SAMMANFATTNING

Nerver för behaglig beröring

I denna avhandling har speciella hudnerver undersökts som tycks vara specialiserade på att skicka nervimpulser till hjärnan när vi blir långsamt smekta över huden. Nervtrådarna kallas CT (C-tactile) och går direkt till områden i hjärnan som är viktiga för uppkomsten av emotioner och reglering av vårt allmänna välbefinnande. Ju effektivare CT-nerverna stimuleras desto behagligare upplever vi att beröringen är. Resultaten kan ha betydelse för förståelse av varför hudberöring kan väcka så starka känslor, varför hudberöring kan bidra till att skapa tillit och förtroende mellan individer och kanske hur hudberöring kan bidra till smärtlindring.

Genom att studera friska försökspersoner med mikroneurografisk teknik, där en tunn elektrod sticks in i en hudnerv för att registrera enstaka nervers signaler, undersöktes hur olika beröringsnervtrådar signalerar vid en mjuk penselstrykning. En datorstyrd robot borstade med en mjuk målarpensel med olika hastighet över det hudområde där den enstaka nervtråden är känslig samtidigt som nervens signaler registrerades. I en parallell studie undersöktes hur behagligt eller obehagligt en grupp friska försökspersoner upplevde penselstrykningen. Det fanns ett starkt samband mellan hur tätt nervsignalerna skickades i CT-nerverna och hur behagligt försökspersonerna uppfattade borststimuleringen. Detta samband var unikt för taktila C-nerver och sågs inte för de någon av de andra typerna av känselnervtrådar vi har i huden.

Vidare undersöktes hur beröring upplevs på hudområden utan CT-nerver. Detta gjordes genom att åter låta försökspersoner gradera behaglighetsupplevelsen av penselstrykningar med varierande hastighet. En grupp patienter undersöktes, som har en genetisk defekt som gör att de har brist på de tunna nerver som utgörs av bland annat taktila C-fibrer. Patienterna uppfattade penselstrykningarna som mindre behagliga jämfört med en ålders- och utbildningsmatchad kontrollgrupp. Ett liknande resultat återfanns när det undersöktes hur lätta penselstrykningar upplevs i handen, där CT-nerver saknas, och jämförde detta med armen som har CT-nerver. När friska försökspersoner blev strukna i handen upplevdes stimuleringen som mindre behaglig i handen jämfört med armen. Resultatet visade även att en serie av tidigare penselstimuleringar på armen kunde påverka hur försökspersoner upplevde en efterföljande stimulering på handen men inte vice versa. Det spekuleras här att sättet på vilket man graderar det emotionella värdet av en hudberöring kan påverkas av tidigare aktivering av CT-nerver och de kretsar i hjärnan som aktiveras av dessa.

Slutlingen användes funktionell magnetresonansavbildning (fMRI) för att undersöka i detalj hur CT-nerver signalerar till hjärnan. Resultatet visade att det system som utgörs av CT-nerver signalerar till delar av hjärnan viktiga för individens välbefinnande och att CT-systemet är organiserat på ett liknande sätt som det system som signalerar smärta och temperatur. Sammantaget visar studierna att CT-nerver signalerar behaglig beröring och är del av ett nätverk för välbefinnande.

LIST OF PUBLICATIONS

This thesis is based on the following papers, which are referred to in the text by their roman numerals.

I. **Löken L.S.**, Wessberg J., Morrison I., McGlone F., Olausson H. Coding of pleasant touch by unmyelinated afferents in humans. *Nature Neuroscience* 2009 May; 12(5): 547-8.

II. Morrison I., **Löken L.S.**, Minde J., Wessberg J., Olausson H. Reduced C afferent fiber density affects perceived pleasantness of touch and empathy for touch. *Manuscript.*

III. **Löken L.S.**, Evert M., Olausson H., Wessberg J. Order effects on affective ratings: pleasantness of touch in hairy and glabrous skin. *Manuscript.*

IV. Björnsdotter M, **Löken L**, Olausson H, Vallbo Å, Wessberg J. Somatotopic organization of gentle touch processing in the posterior insular cortex. *Journal of Neuroscience* 2009, 29(29): 9314-20.

TABLE OF CONTENTS

1 INTRODUCTION

1.1 Pleasant touch

The neural substrates underlying pleasure have recently garnered increased interest. The knowledge gained from research on hedonic mechanisms is suggested to be important for understanding affective disorders (Kringelbach and Berridge, 2009), and the affective aspects of touch have recently been studied and conceptualized (Essick et al., 1999; Francis et al., 1999; Olausson et al., 2002; Rolls et al., 2003; McGlone et al., 2007; Olausson et al., 2008b; Gallace and Spence, 2008; Guest et al., 2009; Lovero et al., 2009; Morrison et al., 2009). Human skin is innervated by a class of slowly conducting unmyelinated afferents, C tactile (CT), responding to innocuous touch. CT afferents have been suggested to have a functional role in signaling pleasant aspects of touch (Vallbo et al., 1999; Olausson et al., 2002). This doctoral thesis includes an electrophysiological examination of CT afferents and takes a psychophysical approach to relate the physiological findings to perception of pleasant touch. Furthermore, the central projection pattern of activity related to signals from CT afferents was explored using functional magnetic resonance imaging.

1.2 Tactile afferents

The somatosensory afferent fiber types innervating the skin can be grouped by their conduction velocities into three classes, from fast to slow: Aβ, Aδ and C-fibers. Nociceptors, itch and temperature fibers are of the C and Aδ subtypes, whereas Aβ are typically described as the afferent type responsible for mediating light touch sensation (Kandel et al., 2000). This thesis is focused on another group of low-threshold mechanoreceptive C-fibers, proposed to signal pleasant touch (Vallbo et al., 1993; Vallbo et al., 1999; Olausson et al., 2002).

1.2.1 Myelinated tactile afferents

Aβ fibers are classically described as mediators of light touch, they have large diameter axons and conduction velocities around 50 m/s in humans. On the basis of adaptation characteristics the Aβ fibers in human hairy skin can be further subdivided into slowly adapting type I (SAI), slowly adapting type II (SAII) and rapidly adapting (RA); hair, field and Pacini (PC) units (Vallbo et al., 1995). Slowly adapting fibers provide a continuous discharge for minutes or more during a constant mechanical stimulation. Rapidly adapting (RA) fibers lack static sensitivity altogether and respond to mechanical changes of stimuli on the skin (Johansson and Vallbo, 1979a, b; Birznieks et al., 2001).

A close relation has been demonstrated between SAI and the perception of texture and form (Johnson et al., 2002). SAII afferents (Ruffini receptors) are particularly sensitive to skin stretch (Vallbo et al., 1995; Olausson et al., 2000). Hair, but not field units, are particularly sensitive to hair movements (Vallbo et al., 1995). The glabrous skin contains four types of mechanoreceptors, SAI, SAII, PC, and RAI (Meissner) units (Johansson and Vallbo, 1979b).

1.2.2 Unmyelinated tactile afferents

Over the past twenty years it has been demonstrated that human hairy skin is also innervated by unmyelinated afferents responding to light touch (CT afferents). CT afferents (in animal research often abbreviated as C low-threshold mechanoreceptors, CLTM) were first described by Zotterman in cats in 1939. The first extensive description of the human homolog of CLTM afferents was achieved in the 1990s (Nordin, 1990), although one such unit had been briefly described two years earlier (Johansson et al., 1988). In humans CT afferents are identified by a low mechanical threshold (< 5 mN) as tested with von Frey monofilaments and slow conduction velocity (1 m/s). CT afferents can produce high-frequency (50-100 impulses/s) trains of action potentials to a gentle, slowly moving stimulus. They have intermediate adaptation properties, responding initially with a burst of high impulse rate, which often falls to zero after a few seconds of sustained indentation (Vallbo et al., 1999). CT afferents may occasionally continue to produce a train of impulses with variable discharge rate after the release of a local skin deformation. This phenomenon is called after-discharge and may last for many seconds. After-discharge is more likely to occur if the stimulus is slowly moving and in conjunction with cooling (Zotterman, 1939; Iggo, 1960; Wiklund Fernström, 2004). Another characteristic of CT afferents is that they are easily fatigued when subjected to repetitive stimuli, and decrease their firing rate when interstimulus intervals are short (Iggo, 1960; Bessou et al., 1971; Iggo and Kornhuber, 1977; Lynn and Carpenter, 1982; Wiklund Fernström, 2004). The time for recovery from fatigue has been reported to range from 30 seconds in humans to up to 30 minutes in cats (Iggo, 1960; Wiklund Fernström, 2004).

CT afferents in humans have been identified in the face and extremities and are likely to be distributed in the hairy skin throughout the body (Nordin, 1990; Vallbo et al., 1993; Vallbo et al., 1999; Edin, 2001; Löken et al., 2007). Numerous recordings have been made from the median nerve, but CT afferents have not been found in glabrous skin. The receptive fields of the CT afferents are round or oval with highly non-uniform terminals (Nordin, 1990; Wiklund Fernström et al.; 1999; Wessberg et al., 2003), which is consistent with animal research indicative of the receptors being free nerve endings (Cauna, 1973, 1976; Liu et al., 2007). Keratinocytes, the predominant cell type in the epidermis, have been proposed as the initial detectors of skin deformation which in turn signal to the sensory afferents' free nerve endings (Denda et al., 2007), although there is currently no (Lumpkin and Caterina, 2007). CT afferents seem to comprise a receptor class carrying unique molecular properties. They show low or negligible activity in response to topical application of capsaicin (2 %), and do not respond to pure thermal stimuli (LaMotte et al., 1992; Wiklund Fernström, 2004). CT afferents are physiologically distinct from nociceptors since the latter do not respond to a soft brush stroking and their response to gentle touch is typically minimal, with only a few impulses at low rate (Vallbo et al., 1999).

CT and myelinated afferents are both sensitive to light mechanical indentations such as soft brush stroking. The ability to code for this event, however, distinguishes the two types. In response to a moving stimulus, myelinated afferents typically show a high dynamic sensitivity, evident as higher impulse rates the faster the stimulus is changing (Bessou et al., 1971; Greenspan, 1992; Edin et al., 1995). Studies of CT units have demonstrated that their response is poor to rapidly moving stimuli, but still they respond strongly to slowly moving stimuli (Bessou et al., 1971; Shea and Perl, 1985; Nordin, 1990; Vallbo et al., 1999). Therefore it seems that CT units fail to code for rapid events, although possessing sensitivity to dynamic stimuli, but only within a low-frequency range.

As CT afferents seem to respond particularly well to moving stimuli, Zotterman proposed early on that CT afferents are likely to account for the sensation of tickle (Zotterman, 1939; Bessou et al., 1971). However, since healthy subjects do not report tickling sensations for stimuli optimal in activating CT afferents, and patients with selective loss of Aβ afferents due to sensory neuronopathy lost their ability to feel tickle when they became ill (Olausson et al., 2002; Olausson et al., 2008a), the tickle hypothesis has now been put aside in favour of a functional role for these afferents in pleasant touch (Vallbo et al., 1993; Vallbo et al., 1999; Olausson et al., 2002). The role of CT afferents in sexual function has not been studied, but studies in mice suggest that they are not present in the genitalia (Liu et al., 2007). In **Paper I** we continued the investigation of how low-threshold mechanoreceptors, and in particular, CT afferents, respond to a dynamic stimulus, such as soft brush stroking, and how this is related to the perception of touch pleasantness.

1.3 Touch pathways

1.3.1 Large-fiber pathway

Tactile impulses travel via Aβ fibers to the ventral posterolateral nucleus of the thalamus (Kandel et al., 2000). From thalamus to cortex, afferents project to the primary and secondary somatosensory cortex (S1, S2) (Maeda et al., 1999), the posterior parietal cortex - specifically, Brodmann's areas 5 and 7 (Mesulam, 1998), and the insular cortex (Schneider et al., 1993). Within S1 and S2 the sensory information from all body surfaces is organized in a somatotopic manner (Penfield and Rasmussen, 1950; Maldjian et al., 1999; Ruben et al., 2001).

1.3.2 Small-fiber pathway

The current view is that CT afferents terminate in the superficial layers of the dorsal horn, in lamina II (Kumazawa and Perl, 1977; Light et al., 1979; Sugiura et al., 1986; Light and Willcockson, 1999), as has been classically described for other small-diameter (Aδ and C) fibers responding to noxious, temperature and itch provoking stimuli (Willis, 1985a, b; Willis and Coggeshall, 1991; Han et al., 1998; Craig et al., 2001; Andrew, 2009), and possibly connect via interneurons to lamina I (Brown and Fyffe, 1981). The lamina I neurons project somatotopically in the spinothalamic pathway in the ventral horn of the spinal cord to the ventral posterior nucleus of the thalamus (in humans, to a distinct area termed the posterior ventromedial nucleus; VMpo; Craig et al., 1994; Dostrovsky and Craig, 1996); for an alternative view see (Willis et al., 2002).

1.4 Cortical processing of CT afferent input

Functional imaging studies in humans suggest the posterior insular cortex as a primary cortical target for C fibers (Olausson et al., 2002; Olausson et al., 2008a), an area strongly interconnected with the amygdala, hypothalamus, orbital frontal cortex, and homeostatic regions of the brainstem (Augustine, 1996; Craig et al., 2000; Craig, 2002; Olausson et al., 2002; Craig, 2003; Olausson et al., 2008a). Mapping of the central neural representation of CT afferent projection has been particularly difficult to obtain as tactile stimulation will always activate myelinated fibers simultaneous with CT fibers in healthy humans. Evidence for CT afferent projection to posterior insula comes from studies of two patients suffering from a rare neuronopathic syndrome, which has left them with no Aβ fiber, but an intact C fiber system. Functional magnetic resonance imaging (fMRI) of soft brush stroking on hairy skin in these patients showed activation in the insular cortex, but no activation in somatosensory cortices (Olausson et al., 2002; Olausson et al., 2008a). In **Paper IV** we further investigated the projection of CT afferents. Specifically, we asked whether CT afferents relay in a somatotopic fashion to the insular cortex as has been shown for other types of C fibers, which signal cooling and pain sensation (Brooks et al., 2005; Hua le et al., 2005; Henderson et al., 2007).

In addition to insular cortex, orbital frontal cortex has been implicated in CT processing (McCabe et al., 2008). The orbital frontal cortex has also been shown to represent painful and pleasant aspects of touch to the palm, demonstrating the relevance of this brain region for representing the emotional dimensions of tactile stimulation as well as the similar networks involved in both painful and pleasant sensation (Francis et al., 1999; Rolls et al., 2003; Kringelbach, 2005; Leknes and Tracey, 2008).

2 AIMS OF THE THESIS

This thesis aspired to answer the following questions:

1. What is the relationship between brush stroking velocity, CT afferent discharge rate, and the perception of pleasantness?

2. How does a selective small-fiber neuropathy affect perception of touch and empathy for touch?

3. What is the relationship between brush stroking velocity and the perception of pleasantness in glabrous skin?

4. Are CT afferent projections to the insular cortex organized somatotopically, as has been shown for other types of C fibers?

3 METHODOLOGICAL CONSIDERATIONS

3.1 Ethics

These studies were approved by the local ethics committee of the medical faculty, University of Gothenburg, Sweden, and experiments were performed according to the Declaration of Helsinki. The declaration emphasizes the subject's right to terminate his or her participation at any time without stating any reason. Informed and written consent was obtained. Reimbursement was provided at SEK 200 per hour.

3.2 Subjects

In **Paper I**, 25 healthy subjects participated in the nerve recordings, and 20 healthy subjects in the psychophysical study. As microneurography recordings demand long experiments (up to about 7-8 hours), special care was taken to explain the subjects' right to terminate their participation in the experiment. The microneurographic recording technique and tactile stimulation was carefully explained to the subject.

In **Paper IV**, 6 healthy subjects and a neuronopathy patient (GL, age 56, right handed, female) participated. This patient suffers from a specific loss of large diameter myelinated fibers (Sterman et al., 1980), leaving unmyelinated afferents intact (Forget and Lamarre, 1995). GL's motor nerve conduction and electromyography are within the range of healthy subjects, and thresholds for temperature and pain detection are largely normal (Olausson et al., 2002; Olausson et al., 2008a). GL denies any ability to identify touch below the level of the face (Forget and Lamarre, 1995). She is, however, able to detect gentle touch to the hairy skin, but not glabrous skin, in a forced choice scenario (Olausson et al., 2002).

In **Paper II**, 10 patients with a rare neuropathy classified as hereditary sensory and autonomic neuropathy type V (HSAN-V), associated with a mutation affecting the neural growth factor beta (NGFB) gene, participated. The patients have normal autonomic and cognitive functions and consider themselves to have normal touch sensibility with no history of allodynia or hyperalgesia. They had normal motor and sensory nerve conduction (except for median nerve compression at the level of the carpal tunnel in three patients) indicating intact function of Aβ fibers, and no other neurological diseases. The patients have, to different degrees, reduced temperature and pain sensations, notably deep pain insensitivity (Minde et al., 2004, 2009) and sural nerve biopsies showed that they had a reduction in C fiber density (Minde et al., 2004). The patients were compared to a group of age, sex and education matched controls (n = 10).

In **Paper III**, 28 healthy subjects participated (10 of these were included in **Paper I**).

3.3 Stimuli and protocols

In **Paper I** and **III** skin stimulation was provided by an artist's flat, soft watercolor brush made of fine, smooth, goat's hair. The width of the brush was 20 mm. In **Paper II** and **IV** gentle stimulation was delivered using a 7 cm wide soft artist's goat hair brush with an indentation force of approximately 0.8 N.

In **Paper I,** and in two of the three experiments in paper **III**, brush strokes were delivered by a robotic device (rotary tactile stimulator, RTS, see below) using velocities of 0.1, 0.3, 1, 3, 10, and 30 cm/s, calibrated normal forces of 0.2 or 0.4 N, and stimulation was made over a 6.5 cm distance.

In **Paper II,** and in one of the three experiments in **Paper III**, brush strokes were delivered manually by an experimenter over a 10 cm distance at 5 different velocities:

0.3, 3, 10, and 30 cm/s. The experimenter was trained in the delivery of the stimuli, and guided by a display on a computer monitor with a stripe moving 10 cm on the screen, representing the appropriate velocity in each trial. The monitor was not visible to the participant.

In **Paper IV**, brush strokes were delivered manually by an experimenter over a 16 cm distance. The brushing velocity varied in the range 4 – 7.5 cm/s.

In **Paper II**, short videos depicting the experimenter stroking another person's skin, using her palm, were randomly intermixed with the tactile stimulus trials. The stroking velocities in the videos were the same as those for tactile stimulation (0.3, 1, 3, 10, and 30 cm/s). The participants were instructed to "rate how pleasant you think the touch feels to the person in the video".

3.4 Rotary Tactile Stimulator (RTS)

In **Paper I** and **III**, a robotic device, known as the RTS was used to deliver brush strokes. The brush was moved perpendicularly to the skin surface in a rotary fashion onto, across, then off the skin by a brushless DC motor (Maxon Motor AG, Sachseln, Switzerland) fitted with a reduction drive and position encoder. A 6-axis force and torque transducer (ATI Industrial Automation, Apex, NC, USA) was mounted between the shaft of the DC motor assembly and the hub, which held a probe and brush. This transducer was as close to the central point of the probe and brush as possible. The DC motor and transducer assembly was mounted on a linear drive, driven by a stepper motor (Parker Hannifin Corp., Rohnert Park, CA, USA). Both the DC and stepper motors were under computer control.

3.5 Statistical considerations

To address the relationship between the different brushing velocities, ratings of pleasantness, and discharge rates in the microneurography recordings, the following test was generally applied: Regression analysis was done by first transforming velocity, the independent variable, to $\log^{10}$ values. To test for significance of a quadratic regression term, the curve fit of a linear regression (reduced regression model) was tested against the fit of a quadratic regression (full regression model), with an F-test for significant reduction of the error sum of squares in the full compared to the reduced model (Chatterjee et al., 2000). When the quadratic regression term provided a significant fit, it is generally described as having an inverted U-shape, which means that the function (here, pleasantness ratings or discharge rates) peaked at intermediate velocities. In addition to the regression model analyses, data was analyzed using standard parametric and non-parametric statistical methods (t-tests, ANOVA, Kruskal-Wallis, Mann-Whitney). All calculations were done in MATLAB (The Mathworks, Natick, MA, USA) or SPSS 15.0 (SPSS Inc., Chicago, IL, USA).

3.6 Microneurography (Paper I)

Recordings from single afferents responding to light touch were obtained using the microneurography technique (Hagbarth and Vallbo, 1967; Vallbo et al., 1979; Vallbo et al., 2004).

3.6.1 Nerve recording and search procedure

Nerve impulses were recorded from single afferents in the lateral antebrachial cutaneous nerve. The nerve, which is a small branch of the musculo-cutaneous nerve, was explored 1-3 cm proximal to the cubital fold. In some cases the dorsal branch of the radial nerve was explored. When the tip of the recording electrode had attained an intrafascicular position, the experimenter searched for single units by lightly stroking his/her fingertips over the skin on the radial surface of the forearm. Any A or C, well-isolated, single unit that readily responded to a brushing stimulus, were further studied.

The nerve signal was recorded using a passive band-pass filter set to 0.2-4.0 kHz. Data was sampled to a PC computer and further analyzed using the ZOOM/SC system developed at the Department of Physiology, Umeå University, Sweden. Sampling rates were 12.8 kHz for the nerve signal, 400 Hz for a strain gauge signal, and 25.6 kHz for timing signals from the RTS that were used to indicate the onset and stop of brush stroking. Each recorded nerve impulse was inspected offline on an expanded time scale, and impulse trains were accepted for subsequent analysis only if they could be properly validated as originating from a single afferent.

3.6.2 Unit identification - exploratory tests

Thresholds to mechanical stimuli were assessed using von Frey monofilament bristles made of nylon wires to give desired forces (0.1, 0.3, 0.6, 1.3, 2.5, 5, 10, 20, 40, 80, and 160 mN). The threshold was determined by finding the bristle of least force able to produce a clear and reproducible response from the unit. Previous studies have shown that it is often difficult to activate the peripheral receptors of CT-afferents electrically (Vallbo et al., 1999); instead, conduction velocity was estimated from the response latency to mechanical taps delivered using a hand-held strain gauge device. The device consisted of a handle carrying a metal bar ending with a Perspex probe with a rounded tip. Strain gauges on the two surfaces of the metal bar were connected to a bridge amplifier to provide a signal of the indentation force. Distinct taps were delivered toward the most sensitive spot within the receptive field, and the latency observed was used for estimating the conduction velocity of the unit (Vallbo et al., 1999).

Myelinated afferents were classified as slowly or rapidly adapting based on the response to a long lasting indentation. SA afferents were further classified as type I or type II, and RA afferents as hair, or field units (for further details on the classification procedure see Vallbo et al., 1999). Unmyelinated afferents (conduction velocity < 2 m/s) were classified as CT if they had monofilament thresholds below 5 mN (Vallbo et al., 1999). Unmyelinated afferents with monofilament thresholds above 5 mN were classified as nociceptors and were not further studied.

3.7 Psychophysics (Papers I – IV)

Subjects were instructed to rate pleasantness using a computerized visual analog scale (VAS) shown on a laptop monitor, with the endpoints unpleasant to pleasant (-10 to 10 in **Papers I** and **III**, and -5 to 5 in **Paper II**), using their right hand. Ratings were performed by dragging a line from the middle of the scale (0) to a chosen position using a computer mouse. Ratings were recorded via subjects' mouse clicks on the scale. After rating, the line returned to neutral position.

In **Paper IV** we performed a two-alternative forced choice test where GL was instructed to report whether the brush stimulation was applied to the right forearm or to the right thigh. Brush strokes were applied manually (see section 3.3) and 32 trials were performed alternatively between forearm and thigh in a pseudo-random order. Visual cues were prevented by a fabric mounted such that the participant could not see the experimenter or brush.

In **Paper II**, an introspective questionnaire, TACTYPE (Deethardt and Hines, 1983) containing 15 items, was used to assess how patients and controls perceive and use interpersonal touch in everyday life. As a test of discriminative functions, in **Paper II**, tactile directional sensibility was tested on the left dorsal forearm using a hand-held stimulator that was moved (speed of 1 cm/s). A forced-choice method was used, and the stimulator was moved over a predetermined distance in either a proximal or distal direction in a pseudorandom order (Norrsell et al., 2001; Löken et al., 2009). The participant was instructed to have his/her eyes closed and verbally report the direction of the movement. The result was summarized in a response profile area (RPA, theoretical range 18–90) that provided a quantitative measure of the directional sensibility of the subject's forearm.

3.8 Functional magnetic resonance imaging (fMRI), Paper IV

fMRI was used to identify the areas activated by brush strokes to the forearm and thigh using a blood oxygen level dependent (BOLD) contrast. The physical basis of the BOLD contrast is oxygenation-dependent magnetic susceptibility of hemoglobin (Bandettini and Ungerleider, 2001). Localized increases in blood flow increase blood oxygenation, making haemoglobin diamagnetic. When hemoglobin is deoxygenated it has paramagnetic properties (Ogawa et al., 1990). These magnetic differences in oxygenated and deoxygenated haemoglobin are used in fMRI as an indicator of neuronal activity (Bandettini and Ungerleider, 2001), although this relationship is not yet thoroughly understood (Logothetis et al., 2001). fMRI has relatively poor temporal resolution (compared to, for example, electroencephalography) as each brain scan (volume) is acquired during 1-4 s. Within each volume, there is a measuring point in a three-dimensional grid with (voxel) dimensions of 2-4 mm, thus providing a good spatial resolution. As the signal change extracted using BOLD is rather low (3-5%), an experimental paradigm usually requires many repeated volume acquisitions.

3.8.1 Experimental paradigm

A block designed paradigm was used, and the stimuli were applied manually according to timing cues from the scanner. All subjects were instructed to focus on the stimulus throughout the experimental session. Three-volume blocks of forearm brushing, thigh brushing, or rest, each with duration of 10.5 s were presented in pseudo-random order. The scanning session consisted of one anatomical and six functional scans. During each functional scan, 13 blocks were obtained in the healthy subjects and 10 in GL.

3.8.2 Data acquisition

Scanning was done with a 1.5 T fMRI scanner (for healthy subjects: Philips Intera; GL: Siemens Sonata). Anatomical scans were acquired using a high-resolution T1-weighted anatomical protocol. Functional scans were acquired using a BOLD protocol and a T2*-weighted gradient-echo, echo-planar imaging (EPI) sequence (healthy subjects: slice thickness 6 mm, in-plane resolution 3.6 x 3.6 mm; GL: slice thickness 4 mm, in-plane resolution 4 x 4 mm).

3.8.3 Preprocessing and general linear model (GLM) analysis

Standard preprocessing steps were applied to the data. All functional data was first slice-time corrected, where shift in acquisition time between the slices in each volume were accounted for to ensure that each voxel represented the same point in time. Next, motion correction was performed, as small movements are inevitable during brain scanning of human subjects (volume three was used as reference for motion correction), and subsequently a filtering for temporal drifts. A general linear model (GLM), where the most activated voxels are identified, was used in the whole-brain analysis and performed on smoothed data. Spatial smoothing is used to provide a degree of spatial integration, in this case using a Gaussian full width at half-maximum filter of 6 mm. A fixed effect model was used to generalize healthy subject activations to the group level. The resulting activation maps were thresholded to a false discovery rate (FDR) of < 0.01.

3.8.4 Multivariate analysis on region of interest

To investigate CT projection patterns in the insular cortex, a multivariate clustering scheme based on pattern recognition concepts was applied (Björnsdotter Åberg and Wessberg, 2009). Standard preprocessing was performed, with the exception of spatial smoothing. The multivariate method aims to identify clusters of voxels where a classifier can maximally differentiate stimulus conditions. Within the contralateral (left) insular cortex, the cluster that maximally differentiated soft brushing from rest was identified in the posterior region in all subjects, consistent with previous observations (Olausson et al., 2002; Olausson et al., 2008a). This was chosen as region of interest for subsequent somatotopy analysis, where the clustering scheme was applied to the forearm/rest and thigh/rest datasets separately. In the current study, the algorithm was used for identifying the one voxel cluster where two conditions are maximally separable, analogous to the GLM most-activated voxels. For classification, linear support vector machines were used (Suykens et al., 2002).

4 SUMMARY OF RESULTS

4.1 Paper I. Relations between afferent activity and ratings of pleasantness

We used a robotic device (RTS) to deliver brush stoking at 6 different velocities (0.1, 0.3, 1, 3, 10, and 30 cm/s) and two calibrated forces (0.2 N and 0.4 N), on the forearm during microneurography recordings as well as during psychophysical sessions.

4.1.1 Primary afferent response to soft brush stroking

We recorded from single afferents in the hairy skin of the forearm responding to innocuous touch (CT, n = 20; myelinated, n = 36).

CT afferents were encountered roughly as often as slowly adapting units. They had lower discharge rates compared to myelinated units. The total numbers of spikes that were elicited by brush stroking were similar for CT, SAI, SAII and hair units, whereas field units responded with fewer impulses. As a measure of reproducibility, the coefficient of variation (standard deviation divided by mean) of the mean firing rate showed no significant differences between the unit classes ($P = 0.76$).

The relationship between brush stroking velocity and mean firing rate was distinctly different between CT and myelinated afferents. CT afferents showed an inverted U-shaped relationship between brushing velocity and mean firing rate with highest responses at 1, 3 and 10 cm/s (Fig. 2a, **Paper I**). In contrast, mean firing rate increased when brushing velocity increased in all myelinated afferent types (Fig. 2d–g, **Paper I**).

In CT afferents, peak firing rate (average peak of the negative quadratic curve) was at 2.1 cm/s. Maximum firing rate showed a brushing velocity dependence similar to mean firing rate for all five unit types. The contact force of the brush had no consistent effect except for SAI units that responded with higher frequency at the higher calibrated force.

4.1.2 Psychophysics

In a separate session, ten subjects rated the pleasantness of the brush stroking on a visual-analog scale (VAS; Fig. 2b, **Paper I**). Regression analysis of brush velocity and VAS scores showed that a negative quadratic regressor provided a better fit than a linear regressor (F-test, $P = 0.036$). Subjects rated 1, 3 and 10 cm/s as being most pleasant and the peak of the fitted quadratic curve was at 2.8 cm/s.

4.1.3 Relation between afferent discharge and perception of pleasantness

Soft brush stroking on hairy skin was perceived as most pleasant when it was delivered at velocities that were most effective at activating CT afferents (1–10 cm/s). There was a significant correlation between mean firing rates and mean ratings of pleasantness for CT units ($P < 0.001$; Fig. 2c, **Paper I**). In contrast, there was no linear relationship between any of the classes of myelinated afferents and pleasantness ratings.

4.2 Paper II. Analyses of patients with reduced number of unmyelinated skin afferents

We investigated how a group of patients classified as suffering from hereditary sensory and autonomic neuropathy type V (HSAN-V), leaving them with reduced C fiber afferent density (n = 10), perceived pleasantness compared to a group of age, sex and education matched healthy controls (n = 10).

4.2.1 Discriminative touch

The tactile directional sensibility test showed that the patients were well within the established normal range (Olausson et al., 1997; Norrsell et al., 2001). This result suggests that C fiber denervated patients have intact discriminative tactile functions.

4.2.2 Pleasant touch

The comparison of patients' mean VAS ratings of pleasantness to the mean ratings of controls' revealed differences between the two groups. For both tactile and video trials, patients rated brush stroking as significantly less pleasant compared to healthy controls (felt touch: $P < 0.001$; observed touch: $P < 0.001$; **Paper II**, Figs. 1 and 2).

Testing for the best model fit (F-test) by adding a quadratic term to the velocity variable, showed that the negative quadratic term did not improve the model for patients ($P = 0.13$), but provided a significantly better fit for controls ($P < 0.001$). The same result was found for ratings of observed touch, where the quadratic term supplied the best fit for controls ($P < 0.001$), but did not improve the model fit for patients ($P = 0.54$). Patients' ratings also had a higher coefficient of variance in their ratings compared to controls ($P < 0.001$, independent samples *t*-test).

The results showed that patients displayed an atypical response to brush stroking at varying velocities, for both ratings of felt touch and observed touch. The control group displayed the expected rating pattern for both felt and observed touch and the difference between groups was most pronounced for stroking velocities optimal for eliciting CT afferent responses (1–3 cm/s).

4.2.3 Relationship between felt and observed touch

Studies on pain as well as touch have suggested that the neural mechanisms enabling our own sensations are the same substrates we draw on to understand pain and touch in others (Keysers et al., 2004; Morrison et al., 2004; Singer et al., 2004). To investigate this relationship in pleasant touch, mean ratings for patients and controls were submitted to a two-way ANOVA with the variables group (patient, control) and modality (tactile, visual). There was a main effect of group ($P < 0.001$), but no effect of modality ($P = 0.21$). There was no interaction between group and modality ($P = 0.95$). The main effect of group indicated differences in rating patterns between patients and controls regardless of modality, and the lack of an effect of modality on ratings indicated that within groups, pleasantness was rated similarly, regardless of touch being felt or merely seen.

4.3 Paper III. Ratings of pleasantness in hairy and glabrous skin in healthy subjects

In a series of psychophysical experiments, we investigated the perception of pleasantness in response to soft brush stroking of varying velocity (0.1 – 30 cm/s) in the glabrous skin of the palm and hairy skin of the arm.

4.3.1 Pleasantness ratings and order of stimulus presentation.

In experiment 1 (n = 10, fig 1, **Paper III**), naive subjects were asked to rate the pleasantness of a series of brush strokes in the palm, which was followed by a short break and then continued with the same series of brush strokes on the arm. In experiment 2 (n = 10, fig 2, **Paper III**), the order of the stimulated skin area was reversed and a series of brush strokes was first applied to the arm, followed by the same protocol on the palm.

In both experiments, pleasantness ratings for arm showed the expected inverted U-shaped relation to brushing velocity, i.e. the regression between pleasantness and velocity was significantly improved by adding a quadratic term ($Ps \leq 0.005$). When the experiment started by stroking the palm, the regression for palm ratings was not significantly improved by adding a quadratic term ($P = 0.232$). In contrast, when brushing started on arm, the palm ratings were similar to arm and the quadratic term did provide a better fit for palm ($P < 0.001$). In fact, pleasantness ratings could only be separated statistically between palm and arm when the experiment had started on the palm (that is, experiment 1).

The difference between palm ratings in the two experiments was most pronounced at 3 cm/s, where they were higher in experiment 2 compared to experiment 1 (independent t-test, $P = 0.042$ (uncorrected)) and significantly lower at 0.1 cm/s in experiment 2 ($P = 0.03$). There was no significant difference at any velocity between arm ratings for experiment 1 and 2. Dividing the pool of data from experiment 1 and 2 into two temporal parts, the first and last showed that there was no significant shift of pleasantness ratings across velocities over time ($P = 0.15$). This indicates that in general, pleasantness ratings neither increased nor decreased over time.

In experiment 3 (n = 8), the arm and palm were alternately stimulated manually in close temporal succession using 5 different velocities (0.3 – 30 cm/s). Here, pleasantness ratings were again better described by adding a negative quadratic term to velocity for both palm and arm ratings (palm, $P = 0.015$; arm, $P = 0.026$), similar to experiment 2. Further, there was no significant difference in palm or arm ratings ($P = 0.304$) (**Paper III**, fig 3).

4.4 Paper IV. Somatotopic projection of CT afferents to insular cortex

A somatototopic organization has been shown in the posterior insular cortex in response to cooling and noxious stimuli, and we here explored whether a similar organization could be seen for CT afferents. We used soft brush stroking on the arm and thigh, during fMRI, in 6 healthy individuals and in a neuronopathy patient (GL) lacking large myelinated fibers.

4.4.1 Whole-brain activations

The patient showed no significant activations in the GLM analysis (table 1, **Paper IV**). There were, however, below-threshold tendencies towards significant activations for both forearm and thigh brushing in the contralateral posterior and mid-insular cortex, consistent with previous observations (Olausson et al., 2002). In healthy controls, on the other hand, there were significant activations in S1 with a clear somatotopic organization (forearm brushing projecting more lateral and inferior than thigh brushing) and bilateral activations in S2 (table 1, **Paper IV**). At the individual level, in 5 of 6 subjects, posterior to mid-insular activations were found confirming previous results (Olausson, et al., 2002).

4.4.2 Insular somatotopy exploration

Using a multivariate analysis, we searched for adjacent still separable activation clusters in response to CT stimulation of the two skin regions. Results showed a somatotopic organization of activations due to forearm and thigh brush stimulation in the posterior insular cortex (Fig. 2, **Paper IV**). The body-map organization with forearm brushing projecting anterior to that of thigh was clearly and consistently identified in all subjects including GL. The subjects' mean distance between cluster centroids was 9.3 mm, and akin to that of GL at 8.9 mm. The mean distance between clusters centroids was maximal in the anterior–posterior (*Y:* 8 mm) plane, whereas the location differences in the remaining planes were nonexistent or small (*X*: 0 mm, *Z*: 4 mm). The patient (GL) did not differ significantly from the norm, clusters being within 2 standard deviations of healthy subjects' values.

4.4.3 Psychophysics

GL could detect whether the arm or thigh was stimulated in 31 out of 32 trials (97% correct), whereas healthy subjects could readily identify the stimulated limb without exception.

5 DISCUSSION

This thesis is focused on the system of unmyelinated afferents which respond to gentle touch of the skin - the tactile C afferents (CT) - and the significance of this system for the perception of pleasantness from gentle touch.

5.1 The pleasant touch hypothesis

For a long time it was assumed that the role of CT afferents in relation to perception was to account for the sensation of tickle (Zotterman, 1939). More recently, an alternative hypothesis was proposed: that an essential role of the CT afferents is to contribute to the pleasantness of the response to gentle touch (Vallbo et al., 1999; Olausson et al., 2002). The pleasant touch hypothesis was suggested on the basis of CT afferent properties. CT afferents are sensitive to innocuous stimulation, especially gentle, slowly moving touch on the skin. They have slow conduction velocity, fatigue easily to repetitive stimuli, and respond poorly to brisk movements (Nordin, 1990; Vallbo et al., 1993; Vallbo et al., 1999; Wessberg et al., 2003). This makes CT afferents suboptimal for a role in encoding the discriminative properties of a stimulus. Crucial support for the pleasant touch hypothesis came from a patient with large fiber neuronopathy, who lacks Aβ afferents but has intact systems of unmyelinated fibers. Psychophysical measurements from this patient revealed that she perceived gentle brush stroking as pleasant (Olausson et al., 2002).

5.2 Psychophysical analysis of pleasantness

It has previously been shown that pleasantness of touch can be psychophysically evaluated with valid and reliable estimates (Essick et al., 1999). In healthy subjects it was found that being stroked on the forearm skin with soft velvet at an intermediate velocity (5 cm/s) was perceived as particularly pleasant, whereas being stroked at slower (0.5 cm/s) or faster velocities (50 cm/s) was less pleasant. Ratings of pleasantness correlated negatively with ratings of unpleasantness, demonstrating their validity (Essick et al., 1999).

We used the visual analogue scale (VAS) scale procedure to assess the attribute of pleasantness of light touch by brush stroking. After a given brush stroke subjects were asked to rate how pleasant (or unpleasant) the particular stroke was. As different velocities were used, a pattern emerged of the relationship between velocity and pleasantness of soft brush strokes. In **Paper I**, we investigated this relationship in the forearm hairy skin using velocities of 0.1 – 30 cm/s. Subjects pleasantness ratings to soft brush stroking of the arm showed an inverted U-shaped pattern with a peak at 1-10 cm/s; brush stroking was rated as most pleasant at intermediary velocities, whereas pleasantness ratings dropped at slow (0.1 and 0.3 cm/s) or fast velocities (30 cm/s).

Similar protocols were used in **Paper I, II** and **III** where the relationship between velocity and pleasantness on hairy skin was highly consistent for groups of healthy subjects. In the hairy skin of C fiber denervated patients (**Paper II**) as well as in the glabrous skin of healthy subjects (**Paper III**) the pattern was distinctly different as will be described in separate sections below.

The VAS procedure has been challenged from a theoretical point of view as a method to explore interindividual differences (Svensson, 2000, 2005). Many factors can likely influence how a single subject rates pleasantness on any given day, for example, the experimental setting, psychological state and traits of the individual. Hence, data from individual subjects should thus be treated with care. However, the VAS seems to provide consistent data when used to analyze the overall pattern of changes in ratings related to stimulus changes (i.e. velocity) on a group level.

5.3 Pleasantness of light touch in relation to response of CT and Aβ afferents

Pioneering studies of correlation between the firing rate of peripheral units and perception was performed by Mountcastle and his co-workers. They found linear relations between neural data from monkey and psychophysical data from humans allowing the conclusions that RAI (Meissner) units are particularly significant for the sensation of low-frequency vibration, whereas Pacini units cover high-frequency components (Mountcastle et al., 1967; Talbot et al., 1968).

Since then a number of correlative psycho-neural analyses have been published, however, our studies are the first to correlate peripheral neural signaling with non-discriminative, hedonic aspects of touch. In **Paper I** we related impulse rate of tactile afferents to subjects estimates of pleasantness of gentle touch. Given the prevailing hypothesis for CT afferents in pleasant touch, we wanted to explore the relationship between CT afferent discharge rate and perception of pleasantness. Sensory input was varied by changing the speed of movement of a light tactile stimulus. We found a significant linear correlation between pleasantness as estimated with VAS and the impulse rate of single CT-afferents. Interestingly, no such linear relation was found for the Aβ afferents. This finding supports the conclusion that the CT system is vital for the construction of the pleasurable aspects of gentle touch.

It may be argued that impulse rate of single afferents may not the most pertinent measure of sensory input. Ideally, the total response of the population should be recorded (Johnson et al., 2002). However, population response is not possible to assess with available methods. Further, it is conceivable that degree of pleasantness might be more dependent on number of impulses in CT afferents elicited by a stimulus than on impulse rate. However, number of impulses is highly dependent on the duration of stimulus, which in turn is closely tied to the velocity of movement in our experimental design. It seems reasonable to assume that, in central structures, the size of the population response of the CT afferents is considered in relation to the duration of the stimulus which is accurately coded by the Aβ afferents.

It seems particularly relevant in relation to the pleasant touch hypothesis that the CT afferents showed a distinctly different profile than the Aβ afferents with regard to velocity of stimulus movement and firing rate, and, further, that the CT profile, but not the Aβ profile, matched the psychophysical estimate of pleasantness as a function of velocity of movement. Although the exact role of the CT system for the construction of a pleasurable sensation of touch has yet to be determined, it seems that the velocity profiles of CT signalling, Aβ signalling, and psychophysical responses are unique and independent pieces of evidence supporting a CT role in the underpinning of pleasant touch sensations.

5.4 Pleasantness of light touch in skin areas lacking CT afferents

5.4.1 Glabrous and hairy skin in healthy subjects

Considering the correlation found between neural discharge rate in CT afferents and pleasantness ratings, we wanted to investigate in detail how pleasantness of touch is perceived on skin sites without CT afferents. In **Paper I**, we found that perception of pleasant touch is indeed different in hairy skin which is CT innervated, and glabrous skin, where CT afferents have never been identified. Comparison of the pleasantness ratings from brush stimulation of palm and arm showed that ratings of brush stimulations of varying velocities were different for these two areas. Specifically, arm ratings showed an inverted U-shaped pattern with a peak at 1–10 cm/s, whereas palm ratings were statistically flat in relation to velocity. This finding fits with the interpretation that pleasantness of touch is dependent on CT afferents in the hairy skin.

5.4.2 Hairy skin with normal and reduced number of unmyelinated afferents

We further investigated how pleasant touch sensations are perceived in patients with C fiber denervation. The patients had a selective degeneration of unmyelinated afferents (HSAN-V). They had a significantly different appreciation of soft brush stroking of varying velocity on the forearm skin compared to age, sex, and education matched healthy controls. The denervated subjects' ratings of pleasantness were lower, especially at 3 cm/s, which is the optimal stimulus velocity for CT afferents. Moreover, the patients' ratings of pleasantness as a function of brushing velocity followed a linear slope rather than the typical inverted U-shaped curve. The results indicate that stimulation of myelinated afferents alone (on hairy skin) is not sufficient to evoke similar pleasant touch sensations across velocities as in healthy subjects. The combination of the findings that ratings of pleasantness differ between normal hairy skin (which is innervated by CT afferents), on the one hand, and hairy skin of C denervated subjects as well as glabrous skin of healthy subjects, on the other, strongly supports the arguments for a specific role of CT afferents in pleasant touch.

5.5 Central projection of CT afferents

Functional imaging studies in humans provide convergent data confirming the role of the posterior insular cortex as a primary cortical target for small fiber systems coding gentle touch, noxious, cooling or itch-provoking stimuli (Craig et al., 2000; Craig, 2002; Olausson et al., 2002; Craig, 2003). A somatotopic organization has been shown for projections of noxious and cooling stimuli in this cortical area (Brooks et al., 2005; Hua le et al., 2005; Henderson et al., 2007). In **Paper IV** we applied soft brush stimulation to the participants' arm and thigh during functional magnetic resonance imaging (fMRI) in order to explore if the CT system exhibit a similar pattern.

As myelinated afferents are always activated to gentle touch concomitantly with CT afferents in healthy subjects, we compared six healthy subjects to a unique patient (GL), lacking Aβ afferents to disentangle cortical activations evoked by input from myelinated (Aβ) afferents and input from CT afferents. In all healthy subjects, as well as in the patient, activations of two adjacent regions in posterior insular cortex were found when gentle touch was applied. Response to stimulation of the forearm was located anterior to that of the thigh. It was concluded that the CT afferent system projects somatotopically to the posterior insular cortex in a similar fashion as previously identified for afferents signalling temperature and pain.

Gentle touch to the hairy skin thus activates the posterior insular cortex, supporting a pathway for CT afferents as projecting alongside thermosensitive and nociceptive afferents to a region implicated in homeostatic control (Olausson et al., 2002; Craig, 2003; Olausson et al., 2008a). The insular cortex is a region of great interest in relation to affective mechanisms, and considered a gateway from sensory systems to the emotional systems of the frontal lobe (Augustine, 1996; Kringelbach 2005; Craig 2009).

The affective pathway formed by CT afferents is perhaps less accessible to conscious self-report, as evidenced by research on the patients (GL and IW), who suffer from a neuronopathy syndrome causing a selective lack of large myelinated afferents (Sterman et al., 1980). Both patients deny touch sensation below the level of the face in daily life but can, in a forced choice task, detect light stroking of a brush. Moreover, GL reports a vague pleasant sensation in response to this stimulus (Olausson et al., 2002). It seems that the somatotopic organization of CT projections to insular cortex fits with the psychophysical finding that neuronopathy patients lacking Aβ afferents have a crude, but certainly not normal, sense of stimulus localization (Olausson et al., 2008a). The somatotopic organization of CT input to insular cortex may be significant for affective-motivational responses; obviously, a gentle touch on the arm, in comparison with the thigh, will have immensely different social implications.

5.6 General discussion

5.6.1 Contextual factors

Hedonic ratings are unlikely to depend solely on bottom-up neural signalling such as a specific peripheral afferents' discharge rate. A host of factors such as the context of the tactile stimulus, expectation, homeostatic state of the individual, culture, gender, and previous experience are highly influential to the perception of pleasantness by means of top-down mechanisms (DiBiase and Gunnoe, 2004; Hertenstein et al., 2006b; Berridge and Kringelbach, 2008).

One such factor was studied in **Paper III**, where we investigated how a preceding exploration of one skin region (forearm or palm) affected responses from the other of the two. The soft brush stroking protocol involved two sessions close in time, either first palm or first forearm stimulation with soft brush stroking, followed by the other. It was found that, when the first session was brushing of the palm, subjects rated these stimuli as less pleasant than brushing on the forearm. Moreover, pleasantness rating as a function of stroking velocity differed: in the hairy skin, but not in the palm, it followed the CT-related inverted U-shaped pattern with a peak at 1–10 cm/s. However, with the reverse order of the two sessions, i.e. first forearm and then palm stimulation, it was found that the ratings were statistically indistinguishable. This was also true when stimuli were applied alternately between forearm and palm in a single session. These results suggest that the perception of pleasantness for palm stimulation is affected by previous stimulation of the arm, but not vice versa. This may signify that central processing differs between these two skin sites. The asymmetrical effect suggests that glabrous skin stimulation carries less affective consequences than hairy skin stimulation.

It may be speculated that a smaller affective impact from palm tactile afferents could be an evolutionary advantage. Prominent affective responses might potentially confound the role of the glabrous skin tactile system for discriminative and motor control functions which are fundamentally important for normal hand functions. Another evolutionary aspect is that the pad skin of non-primate mammals seems to be primarily designed to take the wear and tear in relation to body support and locomotion and less apt to promote socially affective responses.

Since peripheral pathways have only recently been considered in the hedonic domain, the order effect described above might be significant to consider in the design of studies in affective neuroscience in general.

5.6.2 Empathetic aspects of pleasant touch

It has been suggested that we draw on the same neural substrate when we experience our own sensations of touch as when we try to understand the sensations and feelings of another person who we see being touched (Keysers et al., 2004; Morrison et al., 2004). Functional resonance imaging showed that seeing a painful stimulus applied to someone else activates regions that are involved in representing the affective and sensory aspects of our own pain (Morrison et al., 2004; Singer et al., 2004; Jackson et al., 2006; Morrison and Downing, 2007). For the hedonic dimension of touch, observing soft brush stroking to another person's arm activates the same region of posterior insular cortex as feeling the equivalent tactile stimulation (Morrison et al., 2008).

In **Paper II**, HSAN-V patients and controls rated the pleasantness of brush stroking of varying velocity to the forearm. Alternating between tactile stimuli, subjects were asked to estimate the pleasantness of others. To this end, videos depicting caressing on someone else's forearm were shown. The results suggested that both C fiber denervated patients and controls rated observed pleasant touch similar to felt touch, regardless of the differences in rating patterns between the two groups. This finding suggests that the evaluation of others' touch is based on one's own firsthand hedonic-affective experience.

5.6.3 Summary

Findings of the four studies of the present thesis all converge towards the interpretation that the system of unmyelinated tactile afferents (CT) has a fundamentally different functional role than the fast conducting Aβ system. **Paper IV** showed that CT afferents project alongside other small fibers to an area of the brain implicated in homeostatic control and well-being. **Papers I, II,** and **III** provide arguments for the hypothesis that the CT system plays a role in pleasant touch. Particularly, the findings of **Paper I** demonstrate a unique psycho-neural correlate directly supporting the hypothesis. **Paper II** and **III** corroborate the pleasant touch hypothesis by demonstrating, for two separate conditions, that a crucial psychophysical characteristic of CT-innervated skin is failing when cutaneous areas are tested where CT afferents are lacking.

The type of gentle and slowly moving stimulation of hairy skin that effectively activates CT receptors characterizes important and intimate social interactions of pleasurable nature (Morrison et al., 2009), and has been described to communicate love and trust (Hertenstein et al., 2006a). Therefore, it seems that the CT system may be of particular relevance for social and affective aspects of gentle touch, a powerful means of emotional communication.

The role of the CT system in relation to pain mechanisms is an interesting issue that remains largely unexplored. Recently, there have been findings that suggest an interaction that might be relevant for future investigation of approaches aiming at pain relief (Krämer et al., 2007). More knowledge on the neural substrates governing pleasant touch may also be of interest for further understanding certain psychiatric disorders, such as autism, where gentle touch is perceived as unpleasant (Cascio et al., 2007).

6. Conclusions

I. There is a linear relationship between discharge rate in CT afferents and the perception of pleasantness to soft brush stroking.

II. Normal perception of pleasant touch in hairy skin seems to be dependent on CT afferent innervation.

III. We draw on our own sensory experience to understand pleasant touch in others.

IV. Pleasant touch perception in the palm may be influenced by previous experience of CT afferent stimulation.

V. There is a somatotopic organization of gentle touch processing to CT afferent activation in the posterior insular cortex.

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